

## BIOCHEMISTRY

DETECTION OF ENZYME(S) RESPONSIBLE FOR TRANSFORMATION OF OLEIC ACID TO 7, 10-DIHYDROXY-8-OCTADECENOIC ACID FROM *PSEUDOMONAS AERUGINOSA*, WIU-JS. Valerie C. Sershon, Amber. M.Kuhrts<sup>1</sup>, Jenq-Kuen Huang\*, Robert V. Gessner<sup>1</sup>, Kenneth C. Keudell<sup>1</sup>, and Lisa Wen. Departments of Chemistry and <sup>1</sup>Biological Sciences, Western Illinois University, Macomb, Illinois 61455. J-Huang3@wiu.edu

We have previously reported that *Kluyveromyces marxianus* NRRL Y-8281 significantly enhances the biotransformation of oleic acid to 7-hydroxy-8-octadecenoic acid (HOD) and 7,10 dihydroxy-8-octadecenoic acid (DOD) by *Pseudomonas aeruginosa* WIU-JS after co-culturing these microorganisms for 12 and 24 hours, respectively. It has been difficult to isolate the enzyme(s) responsible for such transformation from *P. aeruginosa* WIU-JS because the enzyme(s) is labile to sonification and other means of breaking cells, which is the first step in protein purification. Nevertheless, by using the cell-free extract in the presence or absence of ethylenediamine tetraacetic acid (EDTA), we noted and reported that a divalent metal ion is required for such biotransformation.

We report here that the enzyme (s) responsible for transformation of oleic acid to HOD and DOD by *P. aeruginosa*, WIU-JS is present in an ammonium sulfate fractionation. The cell-free extract, either from *P. aeruginosa* WIU-JS or *K. marxianus* NRRL Y-8281, has been fractionated by ammonium sulfate. By mixing the same ammonium sulfate fraction from individual cell-free extracts for biotransformation we were able to demonstrate that the enzyme activity for such transformation is only present in a certain ammonium sulfate fractionation. However, we observed only a marginal synergistic effect, and less effect of a metal ion on the production of DOD, which is different from our previous observation when the whole cells were co-cultured. Further purification of the enzyme(s) will be undertaken.

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